This is a preprint version. The final published version is available online at https://doi.org/10.1016/j.indcrop.2021.113639

Winter camelina seed quality in different growing environments across Northern America and Europe

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Winter camelina [*Camelina sativa* (L.) Crantz], a multifunctional oilseed crop, offers the potential to sustainably diversify cropping systems across the USA and Europe. However, to promote winter camelina as a wide-spread sustainable and profitable crop, it is imperative to know how different environmental conditions impact its seed oil content and fatty acid (FA) composition. The objective of this study was to compare the seed qualitative traits [i.e., 1000-seed weight (TKW), seed oil content, FA profile and saturation] of a winter camelina cv. Joelle, grown across six different environments (Poland, Italy, Greece, Canada, USA, and Spain). Winter camelina seed qualitative traits varied significantly across environments. Average TKW across regions ranged from 0.77-1.07 g, with the heaviest and the lightest seeds produced in Poland and Spain, respectively. Joelle seed oil content varied across locations from 35.1 to 41.9%. A significant and negative relationship between seed oil content (r^2=0.50, $P \leq 0.05$) and TKW (r^2=0.44, $P \leq 0.05$) and growing degree days (GDD)/number of days from sowing to harvest demonstrated that environments with a short growing cycle and high temperatures depressed seed oil content and seed weight. Joelle seed TKW, oil content, linolenic acid (C18:3) content, and omega-3/omega-6 FA ratio (n-3/n-6) were promoted when grown in environments with prolonged growing seasons and evenly distributed precipitation. Results indicate that growing conditions should be carefully considered for the future large scale production of camelina as prevailing climate variables will likely influence seed quality, thus affecting the suitability for various end-uses.

Keywords: seed weight; oil content; fatty acid composition, precipitation; temperature, environment

Abbreviations: MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids; FA, fatty acid; TKW, thousand kernel weight; GDD, growing degree days: n-3/n-6 = omega-3/omega-6.

1. Introduction

Camelina [*Camelina sativa* (L.) Crantz] is an ancient oilseed crop once commonly cultivated across parts of Europe and Asia (Zubr, 1997; Berti et al., 2016) before being replaced by canola production (*Brassica napus* L. var. oleifera). However, interest in camelina as an oil and protein source for food, feed, and industrial uses has resurged, in part due to its adaptability and fit into modern cropping systems to sustainably intensify crop production while minimizing disruption to food security (Gesch and Archer, 2013; Chen et al., 2015; Leclere at al., 2021; Zanetti et al., 2021). Camelina is a relatively low input crop (Berti et al., 2016) with a short lifecycle that can provide several ecosystem services (Sindelar et al., 2017).

Camelina seeds have a distinct oil composition that is high in heart-healthy polyunsaturated FAs, i.e. linoleic, and α-linolenic acids (55-60% of total FA content), which combined with relevant amounts of eicosenoic acid (11-19%), tocopherols (~700 to 800 $\mu g\ g^{-1}$ oil), and relatively low erucic acid content (<4%) make it well-suited for multiple uses (Berti et al., 2016; Walia et al., 2018; Zanetti et al., 2021). Biodiesel and renewable aircraft fuel meeting ASTM standards have been made from camelina oil (Moser and Vaughn, 2010; Shonnard et al., 2010; Soriano and Narani, 2012), and it also has great potential for manufacturing biopolymers such as plasticizers, lubricants, polyols, adhesives, coatings, resins, and gums (Kim et al., 2015; Zhu et al., 2017). Moreover, camelina seeds are also known to contain about 25 to 34% protein (Campbell et al., 2013; Sintim et al., 2016; Walia et al., 2018), making its meal, together with the oil, as valuable components in livestock and fish diets (Berti et al., 2016; Zanetti et al., 2021).

Genetics and environmental conditions can significantly impact the quantity and quality of seed storage lipids and hence affect the value and use of certain vegetable oils for industrial products and/or food (Singer et al., 2016). Camelina is no exception. Recently, Brock et al. (2020) demonstrated that seed oil content and FA composition significantly differ among *Camelina* species and can substantially differ within the species due to growth conditions, especially temperature. Temperature greatly affects FA synthesis (Singer et al., 2016), and it is common for seed storage lipids to increase in unsaturation under low temperatures and display greater saturation under high temperatures (Linder, 2000). Both temperature and precipitation or water availability, associated with diverse environments, have been implicated in substantial changes in spring camelina seed and oil yields and FA composition (Guy et al., 2014; Obour et al., 2017; Zanetti et al., 2017). For instance, Obour et al. (2017) found that oil content and the proportion of polyunsaturated FAs (PUFA) were greater, and saturated FAs (SFA) and monounsaturated FAs (MUFA) proportions were lower in spring camelina grown at a more northerly latitude in the Great Plains USA compared with a more southerly site. They mainly attributed this to lower temperatures and ample precipitation during flowering and seed development at the northern site. Similarly, in a multi-location trial across Canada and Europe, Zanetti et al. (2017) found that environment rather than genotype affected spring camelina seed yield and quality and also demonstrated that PUFA levels, especially linolenic acid, increased with lower temperatures during seed development and ripening.

Most studies addressing the effects of genotype and environment on camelina seed oil quantity and quality have focused on spring biotypes. Winter biotypes, which differ from spring ones in that they require a period of vernalization to transition to flowering (Anderson et al., 2018), have not received much attention yet. Although there is far fewer winter than spring types that have been identified to date, Gesch et al. (2018) have shown that genetic variation for oil yield and FA composition exists for winter camelina. However, to the best of our knowledge, little research has focused on the impact of diverse environments on seed oil content and quality of winter camelina.

The cultivar Joelle has been the most extensively studied winter camelina genotype. Joelle has proven to perform well as a cash cover crop to diversify Midwest USA cropping systems (Ott et al., 2019) while providing environmental benefits such as scavenging excess soil N to reduce leaching (Weyers et al., 2019), weed suppression (Hoerning et al., 2020), and forage for pollinators (Eberle et al., 2015). Most field research on winter camelina has been focused on the USA's Corn Belt region, where camelina has been identified as a feasible winter cover crop in typical summer annual-winter fallow cropping systems (Wittenberg et al., 2020). Although winter camelina has not been extensively studied in Europe (Kurasiak-Popowska et al., 2018; Zanetti et al., 2020; 2021), spring camelina can be successfully grown with a autumn/winter cycle in Mediterranean areas of Europe (Masella et al., 2014; Royo-Esnal & Valencia-Gredilla, 2018; Righini et al., 2019; Angelini et al., 2020) surviving winter with negligible damage. Thus, to promote winter camelina as a sustainable and profitable crop throughout Europe and North America, it is imperative to know how different environmental conditions in various regions impact its seed oil content and FA composition. This is one of the first studies to compare seed qualitative traits of a winter biotype camelina grown across such a wide variety of environmental conditions. We hypothesize that winter camelina (cv. Joelle) grown in the various regions (Poland, Italy, Greece, Canada, USA, and Spain) will produce seeds with similar oil content and quality. This study aimed to determine the seed oil quality (oil content and fatty acid profile) of winter camelina (cv. Joelle) as affected by variable environmental conditions throughout Europe and North America regions.

2. Materials and methods

2.1 Germplasm and site characterization

The winter camelina cultivar Joelle was used in the present study. It is characterized as winter hardy and relatively high yielding compared with other winter genotypes (Gesch et al., 2018). The seeds of Joelle used at all locations in the study were initially produced in Minnesota and provided by USDA-ARS (Table 1).

The six locations were: two in Northern America (Morris, Minnesota - USA, and Morden, Manitoba - Canada), and four in Europe (Bologna - Italy, Aliartos - Greece, Łężany – Poland, Lleida - Spain). The geographical localization of the test environments permitted to grow Joelle in latitudes between 38°22' N (Aliartos) and 53°58' N (Łężany), and in longitudes ranging from 23°06'E (Aliartos) to -98°06'W (Morden). In addition to covering a wide geographical area, the test locations also cover very different climatic conditions (Table 1), ranging from the south Mediterranean climate of Lleida (Spain) and Aliartos (Greece), which both have a mean annual temperature of 15.2 and 16.7°C, respectively, and mean annual precipitation slightly exceeding 400 mm; to the north Mediterranean climate of Bologna (Italy) with a mean temperature of 13.4°C and cumulative precipitation above 600 mm; to the continental climate of Łężany (Poland) with a mean temperature of 8°C and cumulative precipitation of almost 700 mm; to the cold temperate climate of Morden (Canada) and Morris (USA) with a mean temperature of 3.3°C and 5.8°C, respectively, and cumulative precipitation of 500 and 663 mm, respectively. Furthermore, the test locations were characterized by different soil types, being sandy loam in Aliartos (Greece) and Łężany (Poland), clay loam in Bologna (Italy) and Lleida (Spain), loam in Morden (Canada), and fine loam in Morris (USA) (Table 1).

2.2. Experimental design and cultural practices

Joelle was sown at each location at one or multiple sowing dates between early September 2016 (USA) until mid-January 2018 (Spain). In total, 11 field experiments are included in the present study (Table 2). At each test location, the agronomic management was optimized for camelina, based on previous experience of the crop, local agro-ecosystem variances and available equipment. Joelle seeds were tested for germination prior to sowing of each experiment and found to have > 90% germination rate. The same seeding rate was adopted at all locations (500 seeds m^{-2}), except for at Lleida (Spain) and Morden (Canada) where a higher rate was used corresponding to 800 and 700 seeds m^{-2}, respectively. Row spacing varied with seeding equipment available at each location ranging from 0.15 m up to 0.22 m (Table 2). The previous crop at Morris (USA) was spring wheat (*Triticum aestivum* L.), in Lleida (Spain) was winter barley (*Hordeum vulgare* L.), while at the other locations, it was winter wheat (*Triticum aestivum* L.). Seedbed preparation was carried out at each location with typically available equipment, and apart from Morris (USA) where Joelle was no-tillage seeded, elsewhere cultivation and/or disc harrowing was carried out before sowing (Table 2). The seeding depth was approximately 5-20 mm at all locations, and sowing was carried out mechanically except for in Aliartos (Greece) where it was manual. Plot sizes ranged between 7.5 m^2 in Morden (Canada) to 22.5 m^2 in Lleida (Spain). The fertilization rate was adjusted locally to soil chemical properties and typical camelina agronomic management. The applied fertilization rates are reported in Table 2. Weeds were managed chemically only at Morden (Canada), where both dicot and grass herbicides were sprayed (i.e. trifluralin and quizalofop-P-ethyl) while at all the other locations, only manual weeding was carried out. All experiments were rainfed. The experimental design was a randomized complete block with four replicates, except in Aliartos (Greece) (n=3).

The 50% flowering date was surveyed in some of the trials, as reported in Table 2, following Martinelli and Galasso (2011). At full maturity, Joelle was manually or mechanically harvested, depending on the locally available equipment. Representative seed sub-samples of about 50 g were taken from each plot and cleaned to remove any residual plant parts or external seeds and sent to USDA-ARS, Morris, for qualitative analysis.

2.3 Meteorological data

Meteorological data, including air temperature and precipitation were collected at automated weather stations located on-site or nearby each study site. In particular, daily minimum and maximum temperatures, number of rainy days, and daily precipitation were collected at each site. At all test locations, the accumulated growing degree days from sowing to harvest (GDD; °C d) were calculated [Eq. (1)] using daily maximum air temperature (Tmax), daily minimum air temperature (Tmin) and base temperature (Tbase), for which 4 °C was used for the entire camelina cycle, as suggested by Gesch and Cermak (2011).

$$GDD = \sum[(T_{max}+T_{min})/2 - T_{base}] \qquad \text{Eq. (1)}$$

In the experiments where the 50% flowering date was recorded (Table 2) GDD_{AF} was also calculated for the period from 50% flowering to harvest, adopting the same equation as in Eq. (1).

Furthermore, to better understand the impact of environmental conditions on Joelle seed quality, additional meteorological variables were calculated, as defined in the following equations:

$$GDD/d = \sum[(T_{max}+T_{min})/2 - T_{base}] \text{ / number of d from sowing to harvest} \qquad \text{Eq. (2)}$$

$$\text{Prec/d = cumulative precipitation from sowing to harvest / number of d from sowing to harvest} \qquad \text{Eq. (3)}$$

$$\text{Prec/rainy d = cumulative precipitation from sowing to harvest / number of rainy d} \qquad \text{Eq. (4)}$$

As for GDD, the meteorological variables defined in equations 2 to 4 were also calculated for the period from 50% flowering to harvest (AF), for locations were the 50% flowering date was recorded.

2.4. Laboratory analyses

All qualitative parameters of winter camelina seeds were analyzed in the same laboratory in order to reduce any possible interaction between the analysis techniques and the obtained results. In particular, seed weight and seed oil content were determined at the USDA-ARS laboratory, Morris, (Minnesota, USA) and FA analysis was done at the USDA-ARS laboratory, Peoria, (Illinois, USA).

2.4.1. Seed weight, oil content, and fatty acid profile determination

Seeds were dried to constant weight at 65°C before measuring weight. To determine TKW, three subsamples of 1000 seed per replicated treatment (*n*=12) were counted using an automated seed counter (Series 32669, Seed Processing Holland, Enkhuizen, Netherlands) and then weighed to the nearest tenth of a mg on an analytical balance. The seed oil content was determined on a 5 g of seed sample from each replicated plot using pulsed nuclear magnetic resonance (NMR) (Bruker Minispec mq-10, Bruker, The Woodlands, TX, USA). Prior to measurement, seeds were dried for 3 h at 130 °C and then cooled in a desiccator for 30 min. The NMR was calibrated with pure camelina oil and values of oil contents are reported as a percent.

Fatty acid analyses as methyl esters (FAME) was conducted by gas chromatography (GC) on an Agilent Technologies (Palo Alto, CA, USA) 6890N GC using the methods of Isbell et al. (2015). A standard mix of C8 to C30 saturated FAME GLC (Gas-Liquid Chromatography) mixture supplied by Nu-Check Prep (Elysian, MN, USA) which also contained C18:1, C18:2, C18:3, C20:1, and C22:1 was used to identify retention times of methyl esters. Fatty acid methyl esters were synthesized from oil extracted from approximately 50 camelina seeds per sample as previously described by Isbell et al. (2015).

2.5. Statistical analysis

Prior to ANOVA, the homoscedasticity of variance was verified with Bartlett's Test for $P \leq 0.05$. A one-way ANOVA was adopted to test the effect of the different locations on the seed qualitative traits (i.e., TKW, seed oil content, oleic, linoleic, linolenic, eicosenoic, and erucic acids and the n-3/n-6 ratio). Where different sowing dates were tested, at the same location, sowing date was used as a random effect. When ANOVA revealed statistically different means, the LSD's test was used to separate means ($P \leq 0.05$).

A linear regression study was conducted to understand the effect of the different meteorological variables, for the whole crop cycle and/or for the 50% flowering to harvest period, on the investigated seed qualitative traits of Joelle. When the regression was found significant for $P \leq 0.05$ the coefficient of determination (r^2) was reported. All the statistical analyses were carried using the Statgraphics Centurion 18 software (ver. 18.1.13, Statgraphics Technologies Inc., Virginia, USA).

3. Results

3.1 Weather Conditions

Among all the sites tested, Morden (Canada) was the coldest (3.3°C), whereas Aliartos (Greece) was the warmest (16.7°C) over their 20-year mean annual temperatures (Table 1). Considering the whole camelina growing season, the minimum and maximum temperatures were -11.7 and 26°C, respectively (Table 3), surveyed in Canada and Greece. With respect to precipitation, Łężany (Poland) followed by Morris (USA) were the wettest sites, while Lleida (Spain) was the driest with annual precipitation of 683, 663, and 423 mm, respectively, over their 20-year mean cumulative precipitation and also during camelina growing season with cumulative precipitation of 674, 522, and 247 mm, respectively (Tables 1 and 3). The duration of Joelle crop cycle varied significantly across locations (Table 2), lasting 327 d in Łężany (Poland) in the earlier sowing date and only 148 d in Lleida (Spain). The GDD accumulated from sowing to harvest ranged from 1261 in Morris (USA) to 1758 in Aliartos (Greece) in the earliest sowing dates (Table 2). Furthermore, in the trials where flowering date was determined, the GDD accumulated from 50% flowering to harvest varied greatly, ranging from 565 in Morris (USA), in the earliest sowing date, up to 1066 in Łężany (Poland), in the earliest sowing date (Table 2). The delayed harvest could explain the prolonged after-flowering (AF) phase in Poland due to adverse meteorological conditions observed two weeks before harvest. In all locations, where more than one sowing date was tested, the earlier one always corresponded to the greatest GDD accumulation from sowing to harvest. However, accumulated GDD from 50% flowering to harvest was not influenced by the sowing date.

3.2 Seed Qualitative Traits

The considered seed qualitative traits of Joelle (i.e., TKW, seed oil content, oleic = C18:1, linoleic = C18:2, linolenic = C18:3, eicosenoic = C20:1, erucic = C22:1 contents, n-3/-n6 ratio, MUFA, PUFA, SFA, and PUFA/MUFA ratio) varied significantly among experimental sites (Table 4), but only TKW, C20:1, C22:1, n-3/n-6, and PUFA/MUFA showed a coefficient of variation higher than 10%. The average TKW across all environments varied from 0.77-1.07 g. Across all environments, the heaviest Joelle seeds were produced at Łężany, (Poland, 1.07 g), and the lightest in Morris, (USA, 0.81 g), Aliartos, (Greece, 0.79 g), and Lleida, (Spain, 0.77 g), without significant difference among those three sites (Fig. 1). Joelle seed oil content varied across the test locations from 35.1% to 41.9%. Across environments, Lleida (Spain) produced a significantly lower amount of seed oil followed by Aliartos, Greece; in contrast to four countries (Italy, Poland, USA, and Canada) where seed oil content was significantly higher but comparable among them, with a mean value of ~ 41%.

The FA composition of Joelle varied significantly across test locations (Table 4). The C18:1 content varied from 12.7-14.6%, C18:2 from 14.6-17.8%, C18:3 from 37.1-48.0%, C20:1 from 7.7-14.0%, and C22:1 from 0.8-2.7% (Fig. 2). The 18-carbon chain FA's comprised about 68 to 75% of total oil content with the lowest level at Aliartos (Greece) and the highest at Łężany (Poland). The content of C18:1 was significantly higher at Morden (Canada, 14.6%), Aliartos (Greece, 14.3%), and Bologna (Italy, 14.1%) compared with the other locations; the lowest amount (12.7 %) was found at Łężany (Poland, Fig. 2). The linoleic acid (C18:2) content was significantly higher in Lleida (Spain, 17.8%) as compared to all other locations, whereas it was again the lowest at Łężany (Poland, 14.6%). The content of linolenic (C18:3) was greatest for plants grown at Morden (Canada), reporting a value of 48%, followed by Joelle grown at Łężany (Poland) with a mean value of 40.6% (Fig. 2). In all other environments, the C18:3 content ranged between 37.1% Lleida (Spain) and 38.6% Bologna (Italy) as reported in Figure 2. Also, eicosenoic (C20:1) and erucic acid (C22:1) contents were significantly influenced by growing locations, with Morden (Canada) showing the lowest values for both, corresponding to 7.7% and 0.8%, respectively. In all other growing locations, the C20:1 content was almost double compared to that in Morden (Canada) with a mean value of 13.8% across the other five environments. The erucic acid (C22:1) content was three-fold higher in all other locations (averaging 2.5%) compared with that in Morden (Canada) with small but significant differences between Łężany (Poland) and Lleida (Spain) versus Bologna (Italy), Aliartos (Greece), and Morris (USA) (Fig. 2).

The average contents of MUFAs in Joelle camelina oil varied from 23.5 to 31.1%, PUFAs from 55.5 to 65.3%, SFAs from 8.6 to 10.1%, PUFA/MUFA ratio from 1.79 to 2.8, and n3/n6 ratio from 2.1 to 3.0 (Fig. 3) over the different environments. In Morden (Canada) the contents of PUFA (65.3%), PUFA/MUFA (2.8), and n3/n6 ratios (3.0) were substantially greater than at all other sites. Conversely, contents of MUFA (23.5%) and SFA (8.6%) were considerably lower in Morden (Canada) as compared with other locations (Fig. 3). This demonstrates that camelina produced under the coldest climate (Morden, Canada) increased PUFA content, which constitutes >65% of the total FAs in Joelle oil at the expense of MUFA and SFA (Fig. 3). However, contents of PUFA, MUFA, SFA, PUFA/MUFA, and n3/n6 ratios were found to vary only marginally across the other five locations averaging 56.3%, 30.4%, 9.7%, 1.9%, and 2.4%, respectively (Fig. 3).

To better understand the relationships between Joelle seed qualitative traits and specific meteorological conditions characterizing each experiment (Table 3), a regression study was conducted (Table 5). Earlier studies have reported that Significant regressions ($P \leq 0.05$) were observed among the qualitative seed traits (i.e. TKW, seed oil content, C18:1, C18:2, C18:3, n-3/n-6 ratio, and SFA) and specific meteorological variables. But, for the other seed qualitative traits (C20:1, C22:1, MUFA, PUFA, and PUFA/MUFA ratio), no significant regressions were identified with the studied meteorological variables. Joelle 1000-seed weight (TKW) was significantly ($P \leq 0.05$) and negatively correlated with GDD/d, Prec/rainy d, and Prec $_{AF}$/rainy d $_{AF}$ (Table 5). Likewise, seed oil content was significantly and negatively correlated with GDD/d. Interestingly, none of the variables calculated for the 50% flowering to harvest (AF) period were found significantly correlated to Joelle seed oil content.

Concerning the FA composition, oleic acid (C18:1) content was significantly ($P \leq 0.05$) and negatively related with Prec/rainy d, highlighting that wet seasons (high amount of precipitation over the entire growing season) led to a decrease of this FA (Table 5). The linoleic (C18:2) and linolenic acid (C18:3) contents were positively and negatively related ($P \leq 0.05$) with Prec/rainy d, respectively (Table 5), confirming how these two FAs are inversely related, with C18:3 being directly derived from C18:2 through desaturation. The results also revealed that linoleic acid content was not only positively related ($P \leq 0.05$) with Prec/rainy d from sowing to harvest but also with Prec $_{AF}$/rainy d $_{AF}$. Linoleic acid accumulation in Joelle seeds was enhanced in locations where precipitation was high and concentrated in fewer days.As expected, the n-3/n-6 ratio showed the opposite behavior of that for linoleic acid, increasing in environments where precipitation was lower and more diffused over the entire growing season as well as in AF period. Finally, the saturated fatty acid content (SFA) was found to be positively related ($P \leq 0.05$), similar to linoleic acid, with the meteorological variables linked to precipitation, in particular Prec $_{AF}$/d $_{AF}$, Prec/rainy d, and Prec $_{AF}$/rainy d $_{AF}$ (Table 5).

4. Discussion

Joelle winter camelina was able to grow at all six test environments surviving to very low winter temperatures, as in Morden (Canada), but also to the mild conditions in Lleida (Spain), and Aliartos (Greece), confirming its extensive environmental adaptability. Joelle seed yield varied greatly among test environments ranging from 704 kg DM ha^{-1} in Leida up to 2095 kg DM ha^{-1} in Morden, while seed yield data for Bologna and Morris have already been published by Zanetti et al. (2020).The present study demonstrated how the seed quality of Joelle was highly influenced by growing environment, modifying seed weight, total oil content, and the relative percentages of the main FAs substantially. Nevertheless, it is possible that differences in agronomic management among test locations might have also influenced Joelle seed quality. However, previous studies have shown that winter camelina seed quality traits (e.g., oil content and profile) are more greatly affected by environment than management practices such as seeding rate and tillage (Gesch and Cermak, 2011; Gesch et al., 2018). In the present study, the management used for growing camelina was considered optimal or near optimal for the given region and its soil condition.

Results for seed weight and oil content are in line with the available literature for camelina. In particular, results confirmed that Joelle seed is characterized by relatively small size, as in the study by Wittenberg et al. (2020), who found that the TKW ranged from 0.96-1.28 g across different years and sowing dates in the US Great Plains. However, winter camelina varieties with larger seed size are available, like Bison, but when compared with Joelle, it showed lower winter survival (Gesch et al., 2018), and a study by Canak et al. (2020) indicates that Bison may be more susceptible to drought during germination and emergence. The seed weight of Joelle in the present study increased when precipitation was more evenly distributed throughout the crop's growth cycle, including the 50% flowering to harvest period (Table 4), confirming that camelina is susceptible to heavy moisture conditions causing a general decrease in its qualitative performance (Gesch and Cermak, 2011).

Similar to Gesch et al. (2014), who reported an average seed oil content of Joelle sown in Minnesota, USA, ranging from 38.8 to 42.1%, in the present study, the content was in a comparable range (~ 35-42%). In comparison, studies on spring camelina have shown seed oil content to range from 40 to 44% (Zubr 2003; Waraich et al. 2013). Oil contents in the range of 30 to 40% have also been reported for spring camelina grown under dryland conditions in the US Great Plains (Pavlista et al., 2012; Jiang et al., 2014; Sintim et al., 2016). With respect to Joelle winter camelina, Wittenberg et al. (2020) reported a lower seed oil content (~30%) grown in North Dakota, which was attributed to heat stress that occurred at the seed filling stage. This strongly agrees with our regression analysis that reported a significant negative relation between seed oil content and the GDD/d, explaining how environments characterized by a short growing cycle and high temperatures depressed Joelle seed oil content (i.e. Lleida, Spain, and Aliartos, Greece, in the later sowing date). A similar response to environmental conditions is reported in other winter annual *Brassicaceae* such as oilseed rape (Bouchet et al., 2016) and pennycress (Gesch et al., 2016), confirming that an extended vegetative growth phase allowed plants to accumulate more carbohydrates and nitrogen translating to increased yield.

The significant variation in all the main FAs of Joelle oil reported in the present study differ from the results of Kurasiak-Popowska et al. (2020a), which highlighted the high compositional stability of different winter and spring camelina lines grown in the same environment in Poland over three consecutive growing seasons. However, differences among growing environments in the present study were greater than that in the Poland study.

Even though there was variation among the contents of oleic and linoleic acids across locations, their contents were similar to those reported previously by Gesch and Cermak (2011) for Joelle grown in Minnesota, USA, which averaged 15 and 20%, respectively. These findings suggest that oleic and linoleic acid contents of Joelle seeds remained relatively stable over different environments across Northern America and Europe. Conversely, there was a high variation in the linolenic acid content, in particular between Morden (Canada) and all the other locations, where C18:3 accumulation was about 20% greater (Fig. 2). Being that the expression of the FAD3 desaturase enzyme is promoted by low temperatures at seed filling stages (Rodriguez-Rodriguez, 2013), this likely increased the biosynthesis of linolenic acid at locations like Morden (Canada) and Łężany (Poland), where growing-season temperatures were typically lower, and linolenic acid content was greater (Schulte et al., 2013). Furthermore, in Canada, the contents of eicosenoic and erucic acids were significantly less (7.7 and 0.8%, respectively), presumably in relation to environmental conditions that promoted FAD3 desaturase expression to such an extent that it may have strongly competed with the function of FAE1, which is involved in the elongation of C18:1 to C20:1 and C22:1. Similar results were obtained in mutagenized camelina when the FAE1 activity was genetically blocked (Ozseyhan et al., 2018). Otherwise, among the other locations, the amounts of linolenic, eicosenoic, and erucic acids averaged 38.2, 13.6%, and 2.5%, respectively, similar to values reported previously for Joelle winter camelina (Walia et al., 2018), as well as for a study where spring camelina was grown under a winter cycle (Righini et al. 2019). The concentration of erucic acid across several spring camelina genotypes have been shown to be approximately 3% (Zubr 1997, Zubr 2003; Kirkhus et al., 2013). However, Zubr (1997) provided some of the first evidence that winter-types may tend to have lower erucic content than spring-types. In that study, the mean erucic content was 2.5% across five winter varieties, while that across six spring-types averaged 3%. Moreover, Walia et al. (2018) reported erucic levels as low as 1.1 and

2.0% at physiological maturity of winter camelina grown at two different locations in Minnesota US, which is below or equal to the 2% threshold desired for food-grade oil in the US (Abbott et al., 2003) and 5% in Europe (Council Directive 76/621/EEC, 1976). Also, Kurasiak-Popowska et al. (2020b) confirmed the valuable trait of winter camelina types to accumulate low erucic acid content making their oil a suitable feedstock for multipurpose biobased applications without incurring possible restrictions in their use due to the erucic acid level.

The regression study highlighted significant relationships between the seed qualitative traits studied and the growing environments' meteorological conditions. Interestingly, the valuable and positive qualitative traits of Joelle were negatively related with the same meteorological variables enabling authors to identify environments possibly more suitable for growing winter camelina. A significant and negative relation between seed weight with GDD/d, Prec/rainy d, and Prec $_{AF}$/rainy d $_{AF,}$ indicates that Joelle seed weight was impaired by conditions in which the growing season was short and hot, expressed as GDD/d, and also where precipitation is high and erratic, both along the whole crop cycle and from 50% flowering to harvest (Table 5). The present study also corroborates that TKW, seed oil content, linolenic content, and the n-3/n-6 ratio all increased when Joelle was grown in environments characterized by prolonged growing season and even precipitation distribution. These traits are particularly appreciated by the biobased industry, which is looking for seeds with high weight, high oil content, and increased amounts of linolenic acid and n-3 FAs, fulfilling the needs for producing plasticizers, biopolymers, and biolubricants (Jeon et al. 2019). Likewise, these same traits are being sought by the aquaculture industry (Hixson et al., 2014). Similar to linoleic acid, a positive and significant relation exists between SFA and precipitation, showing that higher rainfall favors the higher SFA content in their oils, which are known to store more energy than unsaturated fatty acids (Linder, 2000).

5. Conclusions

The present results confirm that Joelle winter camelina is highly adaptable, being able to grow across very different environmental conditions in the US Great Plains, Canadian Prairies, Mediterranean Europe, and continental Europe. Winter camelina's seed oil qualitative traits were highly influenced by growing conditions, and this should be carefully considered in the possible future scale-up of winter camelina since some areas might be more suitable than others for specific end-uses related to different FA composition. This response to growing conditions might be further exploited in developing different end products for specific biobased markets, but of course, it should be more carefully considered to geographically scale up winter camelina worldwide.

6. Acknowledgements

As with any study of this magnitude, there were several technicians and students involved in maintaining field plots and collecting data that the authors wish to thank. For the Morris, Minnesota, site we thank Joe Boots, Charles Hennen and Scott Larson for their expert field assistance. For the Bologna, Italy, site we thank Angela Vecchi and Giuseppe Di Girolamo for their supervision in the field trials. For the Morden, Canada, site we thank the Manitoba Pulse & Soybean Growers association for providing funding and Esther Fehr from AgQuest Inc. for conducting the field trial. For Lleida, we thank Francisco Valencia-Gredilla and Joana Serrano for their support in the field work. This research is the result of a study carried out at the University of Warmia and Mazury in Olsztyn, Faculty of Agriculture and Forestry, Department of Genetics, Plant Breeding and Bioresource Engineering, topic number 30.610.007-110. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors. The findings and conclusions in this publication are those of the authors and should not be construed to represent any official USDA or U.S. Government determination or policy.

CRediT authorship contribution statement

Maninder K. Walia: Conceptualization, Data curation; Formal analysis; Writing - original draft; **Federica Zanetti**: Conceptualization; Data curation; Methodology; Software, Formal analysis; Writing - original draft; **Russ W. Gesch**: Conceptualization; Data curation; Software; Supervision; Writing - review & editing; **Michal Krzyżaniak**: Data curation; Writing - review & editing; **Christina Eynck**: Data curation; Writing - review & editing; **Debbie**

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7. References

Abbott, P., Baines, J., Fox, P., Graf, L., Kelly, L., Stanley, G., Tomaska, L., 2003. Review of the regulations for contaminants and natural toxicants. Food Control 14(6), 383–389. https://doi.org/10.1016/S0956-7135(03)00040-9.

Anderson, J.V., Horvath, D.P., Dogramaci, M., Dorn, K.M., Chao, W.S., Watkins, E., Hernandez, A.G., Marks, M.D., Gesch, R.W., 2018. Expression of FLOWERING LOCUS C and a frameshift mutation of this gene on chromosome 20 differentiate a summer and winter annual biotype of *Camelina sativa*. Plant Direct 2(7), e00060. https://doi.org/10.1002/pld3.60

Angelini, L., Abou Chehade, L., Foschi L., Tavarini, S., 2020. Performance and potentiality of camelina (*Camelina sativa* L. Crantz) genotypes in response to sowing date under Mediterranean environment. Agronomy-Basel 10, 1929. https://doi.org/10.3390/agronomy10121929

Berti, M.T., Gesch, R.W., Eynck, C., Anderson, J.V., Cermak, S., 2016. Camelina uses, genetics, production and management. Ind. Crop. Prod. 94, 690-710. https://doi.org/10.1016/j.indcrop.2016.09.034.

Brock, J.R., Scott, T., Lee, A.Y., Mosyakin, S.L., Olsen, K.M., 2020. Interactions between genetics and environment shape *Camelina* seed oil composition. BMC Plant Biol. 20, 423. https://doi.org/10.1186/s12870-020-02641-8.

Bouchet, A.S., Laperche, A., Bissuel-Belaygue, C., Rod Snowdon, R., Nathalie Nesi, N., Stahl, A., 2016. Nitrogen use efficiency in rapeseed. A review. Agron. Sustain. Dev. 36, 38. https://doi.org/10.1007/s13593-016-0371-0

Campbell, M.C., Rossi, A.F., Erskine, W., 2013. Camelina (*Camelina sativa* (L.) Crantz): Agronomic potential in Mediterranean environments and diversity for biofuel and food uses. Crop Pasture Sci. 64(4), 388-398. https://doi.org/10.1071/CP13054

Canak, P., Marjanovic Jeromela, A., Vujoševic, B., Kiprovski, B., Mitrovic, B., Alberghini, B., Facciolla, E., Monti, A., Zanetti, F., 2020. Is drought stress tolerance affected by biotypes and seed size in the emerging oilseed crop camelina? Agronomy-Basel 10, 1856. https://doi.org/10.3390/agronomy10121856.

Chen, C., Bekkerman, A., Afshar, R.K., Neill, K., 2015. Intensification of dryland cropping systems for bio-feedstock production: Evaluation of agronomic and economic benefits of *Camelina sativa*. Ind. Crop. Prod. 71, 114-121. https://doi.org/10.1016/j.indcrop.2015.02.065

Eberle, C.A., Thom, M.D., Nemec, K.T., Forcella, F., Lundgren, J.G., Gesch, R.W., Riedell, W.E., Papiernik, S.K., Wagner, A., Peterson, D.H., 2015. Using pennycress, camelina, and canola cash cover crops to provision pollinators. Ind. Crop. Prod. 75, 20-25. https://doi.org/10.1016/j.indcrop.2015.06.026

Gesch, R.W., Cermak, S., 2011. Sowing date and tillage effects on fall-seeded camelina in the northern Corn Belt. Agron. J. 103, 980–987. https://doi.org/10.2134/agronj2010.0485

Gesch, R.W., Archer, D.W., 2013. Double-cropping with winter camelina in the northern Corn Belt to produce fuel and food. Ind. Crop. Prod. 44, 718-725. https://doi.org/10.1016/j.indcrop.2012.05.023

Gesch, R.W., Archer, D.W., Berti, M.T., 2014. Dual cropping winter camelina with soybean in the northern Corn Belt. Agron. J. 106, 1735–1745. https://doi.org/10.2134/agronj14.0215

Gesch, R.W., Matthees, H.L., Alvarez, A.L., Gardner, R.D., 2018. Winter camelina: crop growth, seed yield and quality response to genotype and sowing rate. Crop Sci. 58, 2089-2098. https://doi.org/10.2135/cropsci2018.01.0018

Gesch, R.W., Royo-Esnal, A., Edo-Tena, E., Recasens, J., Isbell,T.A., Forcella, F., 2016. Growth environment but not seed position on the parent plant affect seed germination of two Thlaspi arvense L. populations. Ind. Crop. Prod. 84, 241-247. http://dx.doi.org/10.1016/j.indcrop.2016.02.006

Guy, S.O., Wysocki, D.J., Schillinger, W.F., Chastain, T.G., Karow, R.S., Garland-Campbell, K., Burke, I.C., 2014. Camelina: Adaptation and performance of genotypes. Field Crop. Res. 155, 224-232. https://doi.org/10.1016/j.fcr.2013.09.002

Hixson, S.M., Parrish, C.C., Anderson, D.M., 2014. Full substitution of fish oil with camelina (*Camelina sativa*) oil, with partial substitution of fish meal with camelina meal, in diets for farmed Atlantic salmon (*Salmo salar*) and its effect on tissue lipids and sensory quality. Food Chem. 157, 51-61. doi: 10.1016/j.foodchem.2014.02.026.

Hoerning, C.A., Wyse, D.L., Wells, M.S., Gesch, R.W., Forcella, F., 2020. Yield tradeoffs and weed suppression in a winter annual oilseed relay-cropping system. Agron. J. 112, 2485-2495. doi: 10.1002/agj2.20160

Isbell, T.A., Cermak, S.C., Dierig, D.A., Eller, F.J., Marek, L.F., 2015. Registration of Katelyn *Thlaspi arvense* L. (Pennycress) with improved non dormant traits. J. Plant Reg. 9, 212–215.

Jeon, J.Y., Han, Y., Kim, Y.W., Lee, Y.W., Hong, S., Hwang, I.T., 2019. Feasibility of unsaturated fatty acid feedstocks as green alternatives in bio-oil refinery. Biofuels Bioprod. Biorefining 13(3), 690-722. https://doi.org/10.1002/bbb.1979

Jiang, Y., Caldwell, C.D., Falk, K.C., 2014. Camelina seed quality in response to applied nitrogen, genotype and environment. Can. J. Plant Sci. 94, 971-980. https://doi.org/10.4141/cjps2013-396

Kim, N., Li, Y., Sun, X.S., 2015. Epoxidation of *Camelina sativa* oil and peel adhesion properties. Ind. Crop. Prod. 64, 1–8. https://doi.org/10.1016/ j.indcrop.2014.10.025

Kirkhus, B., Lundon, A.R., Haugen, J.-E., Vogt, G., Borge, G.I.A., Henriksen, B.I.F., 2013. Effects of environmental factors on edible oil quality of organically grown Camelina sativa. J. Agric. Food Chem. 61, 3179-3185. DOI: 10.1021/jf304532u.

Kurasiak-Popowska, D., Graczyk, M., Przybylska-Balcerek, A., Stuper-Szablewska, K., 2020a. Influence of variety and weather conditions on fatty acid composition of winter and spring *Camelina sativa* varieties in Poland. Eur. Food Res. Technol. https://doi.org/10.1007/s00217-020-03639-0

Kurasiak-Popowska, D., Graczyk, M., Stuper-Szablewska, K., 2020b. Winter camelina seeds as a raw material for the production of erucic acid free oil. Food Chem. 330, 127265. https://doi.org/10.1016/j.foodchem.2020.127265.

Kurasiak-Popowska, D., Tomkowiak, A., Czlopińska, M., Bocianowski, J., Weigt, D., Nawracała, J., 2018. Analysis of yield and genetic similarity of Polish and Ukrainian *Camelina sativa* genotypes. Ind. Crop. Prod. 123, 667-675. https://doi.org/10.1016/j.indcrop.2018.07.001

Leclère, M,. Lorent, A.R., Jeuffroy, M.H., Butier, A., Chatain, C., Loyce, C., 2021. Diagnosis of camelina seed yield and quality across an on-farm experimental network. Eur. J. Agron. 122, 126190. https://doi.org/10.1016/j.eja.2020.126190

Linder, C.R., 2000. Adaptive evolution of seed oils in plants: accounting for the biogeographic distribution of saturated and unsaturated fatty acids in seed oils. Am. Nat. 156:442–458. doi: 10.1086/303399

Martinelli, T., Galasso, I., 2011. Phenological growth stages of *Camelina sativa* according to the extended BBCH scale. Ann. Appl. Biol., 158(1), 87–94. https://doi.org/10.1111/j.1744-7348.2010.00444.x

Masella, P., Martinelli, T., Galasso, I., 2014. Agronomic evaluation and phenotypic plasticity of *Camelina sativa* growing in Lombardia, Italy. Crop Pasture Sci. 65(5), 453-460. https://doi.org/10.1071/CP14025

Moser, B.R., Vaughn, S.F., 2010. Evaluation of alkyl esters from *Camelina sativa* oil as biodiesel and as blend components in ultralow-sulfur diesel fuel. Bioresour. Technol. 101, 646-653. https://doi.org/10.1016/j.biortech.2009.08.054

Obour, A.K., Obeng, E., Mohammed, Y.A., Ciampitti, I.A., Durrett, T.P., Aznar-Moreno, J.A., Chen, C., 2017. Camelina seed yield and fatty acids as influenced by genotype and environment. Agron. J. 109, 947-956. https://doi.org/10.2134/agronj2016.05.0256

Ott, M.A., Eberle, C.A., Thom, M.D., Archer, D.W., Forcella, F., Gesch, R.W., Wyse, D.L., 2019. Economics and agronomics of relay-cropping pennycress and camelina with soybean in Minnesota. Agron. J. 111, 1281-1292. https://doi.org/10.2134/agronj2018.04.0277

Ozseyhan, M.E., Kang, J., Mu, X., Lu, C., 2018. Mutagenesis of the FAE1 genes significantly changes fatty acid composition in seeds of *Camelina sativa*. Plant Physiol. Biochem. 123, 1-7. DOI:10.1016/j.plaphy.2017.11.021

Pavlista, A.D., Baltensperger, D.D., Isbell, T.A., Hergert, G.W., 2012. Comparative growth of spring-planted canola, brown mustard and camelina. Ind. Crop. Prod. 36, 9–13. https://doi.org/10.1016/j.indcrop.2011.07.020

Righini, D., Zanetti, F., Martinez-Force, E., Mandrioli, M., Toschi, T G., Monti, A., 2019. Shifting sowing of camelina from spring to autumn enhances the oil quality for bio-based applications in response to temperature and seed carbon stock. Ind. Crop. Prod. 137, 66-73. https://doi.org/10.1016/j.indcrop.2019.05.009

Rodríguez-Rodríguez, M.F., Sánchez-García, A., Salas, J.J., Garcés, R., Martínez-Force, E., 2013. Characterization of the morphological changes and fatty acid profile of developing *Camelina sativa* seeds. Ind. Crop. Prod. 50, 673–679. https://doi.org/10.1016/j.indcrop.2013.07.042

Royo-Esnal, A., Valencia-Gredilla, F., 2018. Camelina as a rotation crop for weed control in organic farming in semiarid Mediterranean climate. Agric. Basel 8, 156. https://doi.org/10.3390/agriculture8100156

Schulte, L.R., Ballard, T., Samarakoon, T., Yao, L., Vadlani, P., Staggenborg, S., Rezac, M., 2013. Increased growing temperature reduces content of polyunsaturated fatty acids in four oilseed crops. Ind. Crop. Prod. 51, 212-219. https://doi.org/10.1016/j.indcrop.2013.08.075

Shonnard, D.R., Williams, L., Kalnes, T.N., 2010. Camelina-derived jet fuel and diesel: sustainable advanced biofuels. Environ. Prog. Sustain. Energy 3, 382-392. https://doi.org/10.1002/ep.10461

Sindelar, A., Schmer, M., Gesch, R.W., Forcella, F., Eberle, C.A., Thom, M., Archer, D.W., 2017. Winter oilseed production for biofuel in the U.S. Corn Belt: Opportunities and limitations. GCB Bioenergy 9, 508-524. https://doi.org/10.1111/gcbb.12297

Singer, S.D., Zou, J., Weselake, R.J., 2016. Abiotic factors influence plant storage lipid accumulation and composition. Plant Sci. 243, 1–9. doi: 10.1016/j.plantsci.2015.11.003.

Sintim, H.Y., Zheljazkov, V.D., Obour, A.K., Garcia, Y., Garcia, A., Foulke, T.K., 2016. Evaluating agronomic responses of camelina to seeding date under rain-fed conditions. Agron. J. 108, 349-357. https://doi.org/10.2134/agronj2015.0153

Soriano, N.E., Narani, A., 2012. Evaluation of biodiesel derived from *Camelina sativa* oil. J Am Oil Chem Soc. 89(5), 917-923. https://doi.org/10.1007/s11746-011-1970-1

Walia, M.K., Wells, M.S., Cubins, J., Wyse, D., Gardner, R.D., Forcella, F., Gesch, R.W., 2018. Winter camelina seed yield and quality responses to harvest time. Ind. Crop. Prod. 124(5), 765-775. https://doi.org/10.1016/j.indcrop.2018.08.025

Waraich, E.A., Ahmed, Z., Ahmad, R., Ashraf, M.Y., Saifullah, Naeem, M.S., Rengel, Z., 2013. *Camelina sativa*, a climate proof crop, has high nutritive value and multiple-uses: a review. Aust. J. Crop Sci. 7(10), 1551-1559.

Weyers, S.L., Thom, M.D., Forcella, F., Eberle, C.A., Matthees, H.L., Gesch, R.W., Ott, M., Feyereisen, G.W., Strock, J.S., Wyse, D., 2019. Potential for nutrient loss reduction in winter oilseed cover cropped systems in the Upper Midwest. J. Environ. Qual. 48, 660-669. https://doi.org/10.2134/jeq2018.09.0350

Wittenberg, A., Anderson, J.V., Berti, M.T., 2020. Crop growth and productivity of winter camelina in response to sowing date in the northwestern Corn Belt of the USA. Ind. Crop. Prod. 158, 113036. https://doi.org/10.1016/j.indcrop.2020.113036

Zanetti, F., Eynck, C., Christou, M., Krzyżaniak, M., Righini, D., Alexopoulou, E., Stolarski, M.J., Van Loo, E.N., Puttick, D., Monti, A., 2017. Agronomic performance and seed quality attributes of camelina (*Camelina sativa* L. Crantz) in multi-environment trials across Europe and Canada. Ind. Crop. Prod. 107, 602-608. https://doi.org/10.1016/j.indcrop.2017.06.022

Zanetti, F., Gesch, R.W., Walia, M.K., Johnson, J.M.F., Monti, A., 2020. Winter camelina root characteristics and yield performance under contrasting environmental conditions. Field Crop. Res. 252, 107794. https://doi.org/10.1016/j.fcr.2020.107794

Zanetti, F., Alberghini B., Marjanović Jeromela, A., Grahovac, N., Rajković, D., Kiprovski, B., Monti, A., 2021. Camelina, an ancient oilseed crop actively contributing to the rural renaissance in Europe. A review. Agron. Sustain. Develop. (2021) 41:2. https://doi.org/10.1007/s13593-020-00663-y

Zhu, X., Wang, D., Li, N., Sun, S.S., 2017. Bio-based wood adhesive from camelina protein (a biodiesel residue) and depolymerized lignin with improved water resistance. ACS Omega 2, 11, 7996–8004. https://doi.org/10.1021/acsomega.7b01093.

Zubr, J., 1997. Oilseed crop: *Camelina sativa*. Ind. Crop. Prod. 6, 113-119. https://doi.org/10.1016/S0926-6690(96)00203-8.

Zubr, J., 2003. Qualitative variation of *Camelina sativa* seed from different locations. Ind. Crop Prod. 17(3), 161-169. https://doi.org/10.1016/S0926-6690(02)00091-2.

Figure captions

Figure 1. The thousand-seed weight (g) and total seed oil content (%) of Joelle camelina grown at the six test locations. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, *P*≤0.05).

Figure 2. Main FAs (C18:1 = oleic acid, C18:2 = linoleic acid, C18:3 = linolenic acid, C20:1= eicosenoic acid, C22:1= erucic acid) characterizing the oil of Joelle camelina grown at the six test locations. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, *P*≤0.05).

Figure 3. Unsaturated FA contents (MUFA and PUFA), PUFA/MUFA ratio, n-3/n-6 ratio (ratio between omega-3 and omega-6 FAs), and Saturated (SFA) of Joelle camelina grown at each test location. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, *P*≤0.05).

Table 1. Country, location, soil type, and main climatic characteristics (20-year historical data) of the six study sites.

Country	Location	Coordinates	Soil type	Mean annual precipitation (mm)	Mean annual temperature (°C)
USA	Morris	45° 35' N, -95° 54' W	Fine Loam	663	5.8
Canada	Morden	49° 11’ N, -98° 06’ W	Loam	500	3.3
Italy	Bologna	44° 33’ N, 11° 23’ E	Clay Loam	613	13.4
Greece	Aliartos	38° 22’ N, 23° 06’ E	Sandy Loam	485	16.7
Poland	Łężany	53° 58’ N, 21° 09’ E	Sandy Loam	683	8.0
Spain	Lleida	41° 37’ N, 0° 37’ E	Clay Loam	423	15.2

Table 2. Sowing, 50% flowering (50% F) and harvesting dates, cycle duration, GDD (Growing Degree Days) from sowing to harvest, GDD_{AF} (from 50% flowering to harvest), and the main agronomic practices adopted in the 11 experiments with Joelle across six locations in Europe, Canada, and USA. Sowing and harvest years were 2016 and 2017, respectively for all locations, except Spain where 2018 the year for both sowing and harvest.

Country	Location	Sowing	Harvest	50% F#	Cycle	GDD*	GDD_{AF}*	Seeding rate	Row spacing	Tillage	NPK
			Date		(d)			(seeds m^{-2})	(m)		(kg ha^{-1})
USA	Morris	6 Sept	30 Jun	19 May	297	1473	565	500	0.19	None	78-34-34
		4 Oct	07 Jul	28 May	276	1261	625				
Canada	Morden	21 Sept	24 Jul	25 May	306	1359	837	700	0.22	Disk + harrow	67-44-0
Italy	Bologna	13 Oct	29 May	02 Apr	228	1360	679	500	0.13	Plough + rotary till	50-83-0
		25 Oct	31 May	07 Apr	218	1290	662				
Greece	Aliartos	27 Oct	31 May	22 Mar	216	1758	949	500	0.15	Plough + rotary till	83-45-45
		17 Nov	06 Jun	04 Apr	201	1651	967				
		13 Dec	10 Jun	17 Apr	179	1583	907				
Poland	Łężany	09 Sept	02 Aug	12 May	327	1603	1066	500	0.15	Plough +rotary till +	30-14-30
		29 Sept	09 Aug	N/A	314	1501	N/A			harrow + roll	
Spain	Lleida	16 Jan	13 Jun	N/A	148	1322	N/A	800	0.18	Plough+ rotary till	0-0-0

N/A= data not available

[#]Flowering year was 2017 for all locations, except Spain where it was in 2018.

*Tbase for GDD calculation 4°C (Gesch and Cermak, 2011)

Table 3. Mean monthly air temperature and accumulated precipitation during the study period at each location.

Months	Morris (USA)*		Morden (Canada)*		Bologna (Italy)*		Aliartos (Greece)*		Łężany (Poland)*		Lleida (Spain)**	
	Mean temp (°C)	Total ppt (mm)	Mean temp (°C)	Total ppt (mm)	Mean temp (°C)	Total ppt (mm)	Mean temp (°C)	Total ppt (mm)	Mean temp (°C)	Total ppt (mm)	Mean temp (°C)	Total ppt (mm)
September	16.7	42.9	14.4	89.8	-	-	-	-	14.7	46.2	-	-
October	9.6	87.1	6.9	28.4	13.5	103.6	18.1	21.0	6.6	151.4	-	-
November	4.8	42.2	4.2	32.9	9.2	83.6	13.2	25.5	2.4	86.2	-	-
December	-9.2	32.5	-11.7	41.0	4.5	32.2	6.8	31.0	1.2	48.1	-	-
January	-9.1	13.2	-10.7	17.8	1.3	5.0	4.7	55.7	-3.2	17.4	7.9	24.6
February	-2.8	11.4	-7.9	13.0	6.5	51.2	9.9	20.7	-1.2	34.0	5.9	30.2
March	-0.8	11.7	-5.1	14.0	11.6	9.6	11.9	71.6	4.9	60.0	10.5	32.4
April	7.6	65.0	5.4	7.9	14.2	33.4	15.5	21.3	7.0	53.8	14.7	69.8
May	13.5	92.5	12.4	21.9	18.3	55.8	21.4	111.6	13.3	19.8	18.5	63.0
June	19.8	101.1	17.9	64.7	-	-	26.2	38.2	16.7	67.0	22.9	14.9
July	22	22.6	19.7	38.3	-	-	-	-	17.7	90.4	26.0	12.1
Avg/Cumulative	6.6	522.2	4.1	369.7	9.9	374.4	14.2	396.6	7.3	674.3	15.2	247.0

* refer to 2016/17 Joelle growing cycle

**refer to 2018 Joelle growing cycle

Table 4. ANOVA results (F-value) for all the considered camelina seed qualitative traits (i.e. TKW = 1000-seed weight, C18:1 = oleic acid, C18:2 = linoleic acid, C18:3 = linolenic acid, C20:1= eicosenoic acid, C22:1= erucic acid, n-3/n-6 = ratio between omega 3 and omega 6 FAs, MUFA = monounsaturated fatty acids, PUFA= polyunsaturated fatty acids, SFA = saturated fatty acids) and the coefficient of variation (CV) in the Joelle camelina study across six study locations.

Factors	Location[1]	Significance	CV(%)
TKW	71.12	***	13.6
Oil content	16.91	***	5.86
C18:1	18.93	***	5.38
C18:2	36.47	***	6.92
C18:3	126.28	***	8.40
C20:1	111.26	***	14.4
C22:1	116.60	***	22.5
n-3/n-6	106.32	***	12.7
MUFA	83.71	***	7.6
PUFA	78.23	***	5.0
SFA	71.90	***	5.9
PUFA/MUFA	66.12	***	15.7

[1]At Morris (USA), Bologna (Italy), Aliartos (Greece) and Lezeny (Poland) multiple sowing dates have been considered.

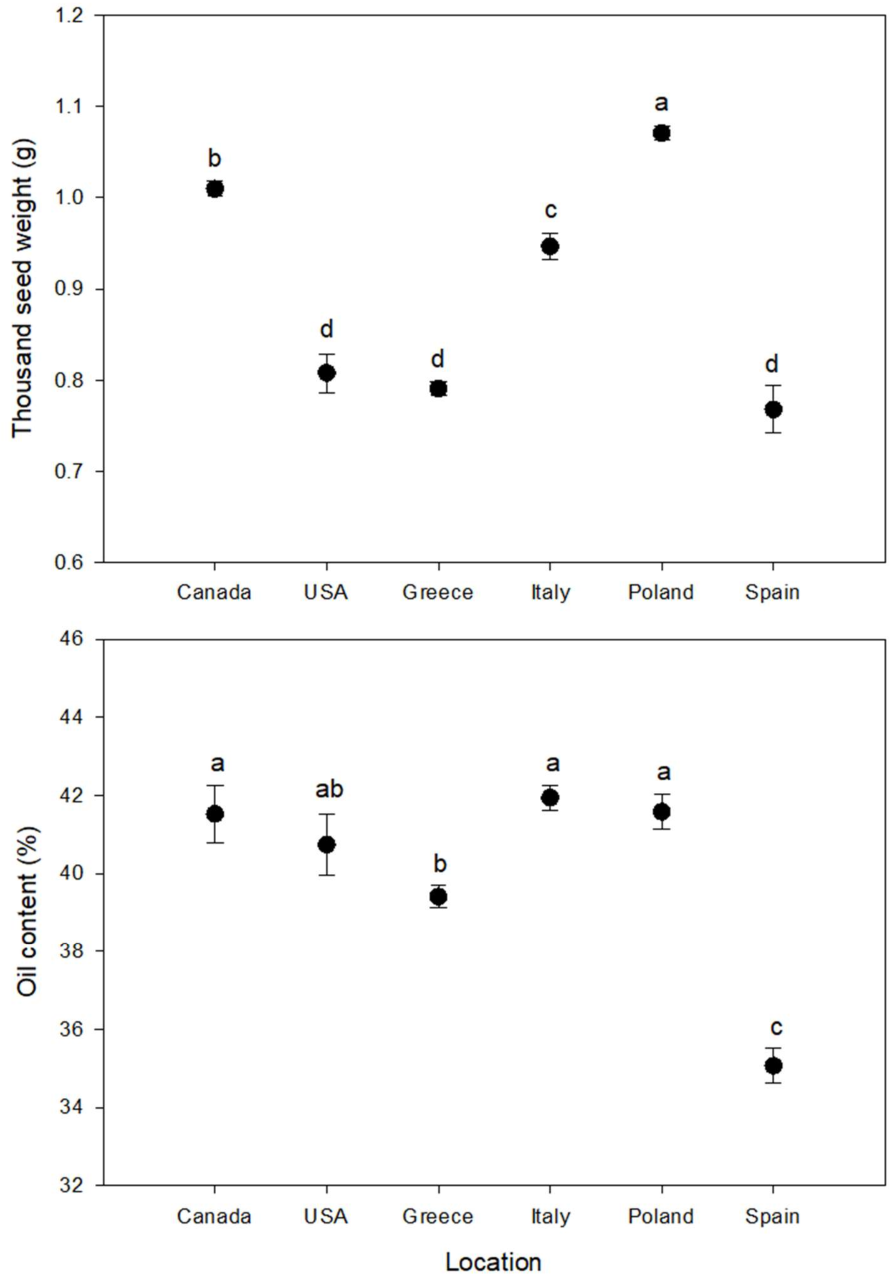
*** Significant at 0.001 probability level.

Table 5. Coefficients of determination (r^2) and *P*-values (*in parenthesis*) for the significant linear regressions between the meteorological variables, calculated for the entire crop cycle and for the 50% flowering to harvest (AF) period, and Joelle seed qualitative traits.

Variables	GDD/d	Prec/d	Prec $_{AF}$/d $_{AF}$	Prec/rainy d	Prec $_{AF}$/rainy d $_{AF}$
TKW	0.44 (*0.025*)	-	-	0.86 *(<0.001)*	0.88 *(<0.001)*
Oil content	0.50 *(0.016)*	-	-	-	-
C18:1	-	0.52 *(0.013)*	-	-	-
C18:2	-	-	0.53 *(0.026)*	0.56 *(0.008)*	0.70 *(0.005)*
C18:3	-	-	-	0.37 *(0.048)*	-
n-3/n-6	-	-	-	0.65 *(0.003)*	0.71 *(0.004)*
SFA	-	-	0.55 *(0.023)*	0.74 *(<0.001)*	0.87 *(<0.001)*

Qualitative traits: TKW = 1000-seed weight, C18:1 = oleic acid, C18:2 = linoleic acid, C18:3 = linolenic acid, n-3/n-6 = ratio between omega 3 and omega 6 FAs, SFA = saturated fatty acids.

Meterological variables: GDD/d = Growing degree days/number od d from sowing to harvest, Prec/d= cumulative precipitation from sowing to harvest / number of d from sowing to harvest, Prec $_{AF}$/d $_{AF}$= cumulative precipitation from 50% flowering to harvest / number of d from 50% flowering to harvest, Prec/rainy d = cumulative precipitation from sowing to harvest / number of rainy d, Prec $_{AF}$/rainy d $_{AF}$= cumulative precipitation from 50% flowering to harvest / number of rainy d from 50% flowering to harvest.



Thousand seed weight (g)
1.2
1.1
1.0
0.9
0.8
0.7
0.6
b
d
d
c
a
d
Canada
USA
Greece
Italy
Poland
Spain
Oil content (%)
46
44
42
40
38
36
34
32
a
ab
b
a
a
c
Canada
USA
Greece
Italy
Poland
Spain
Location

Figure 1. The thousand-seed weight (g) and total seed oil content (%) of Joelle camelina grown at the six test locations. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, $P \leq 0.05$).

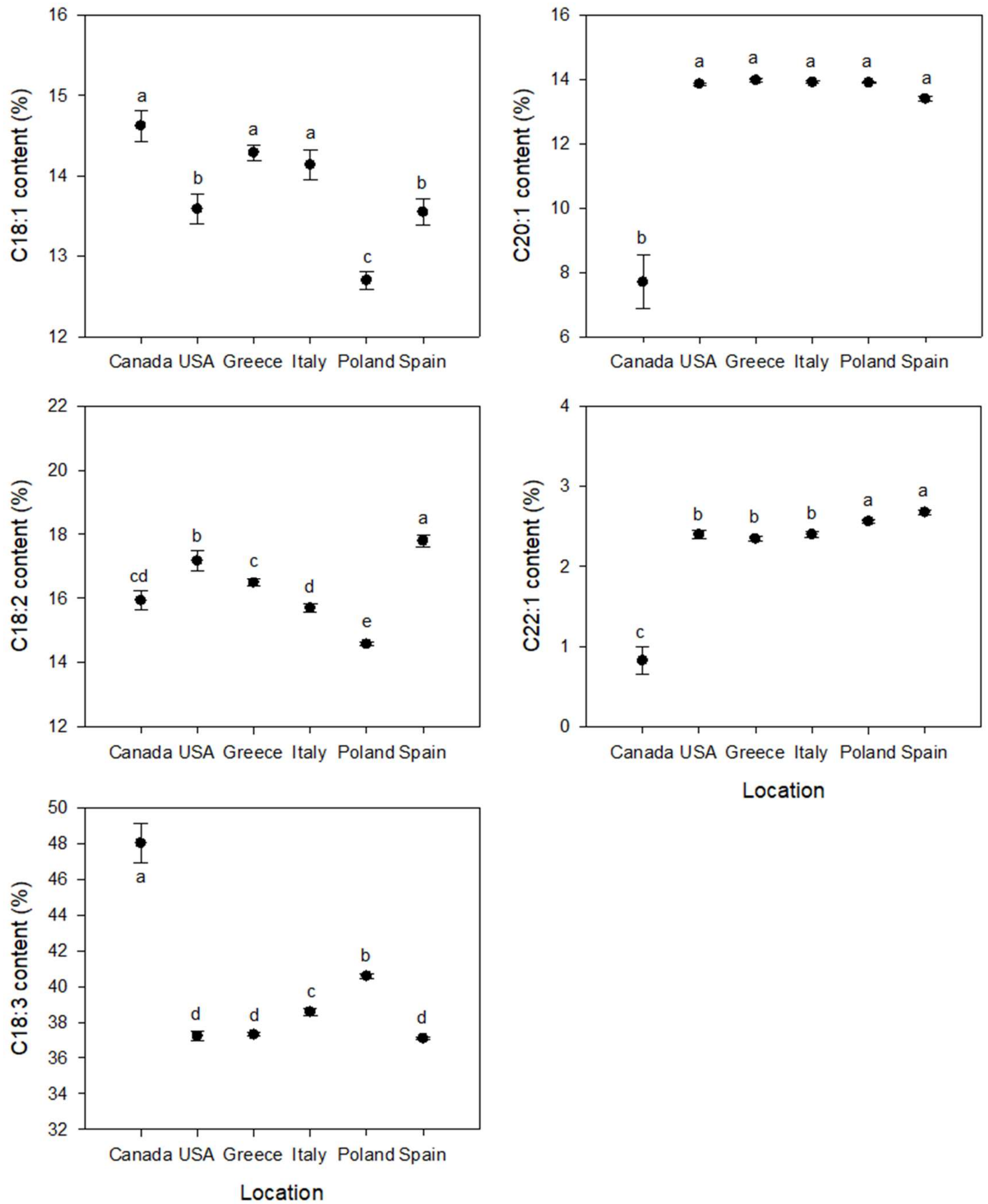

C18:1 content (%)
C20:1 content (%)
C18:2 content (%)
C22:1 content (%)
C18:3 content (%)
Canada USA Greece Italy Poland Spain
Location

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Figure 2. Main FAs (C18:1 = oleic acid, C18:2 = linoleic acid, C18:3 = linolenic acid, C20:1= eicosenoic acid, C22:1= erucic acid) characterizing the oil of Joelle camelina grown at the six test locations. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, *P*≤0.05).

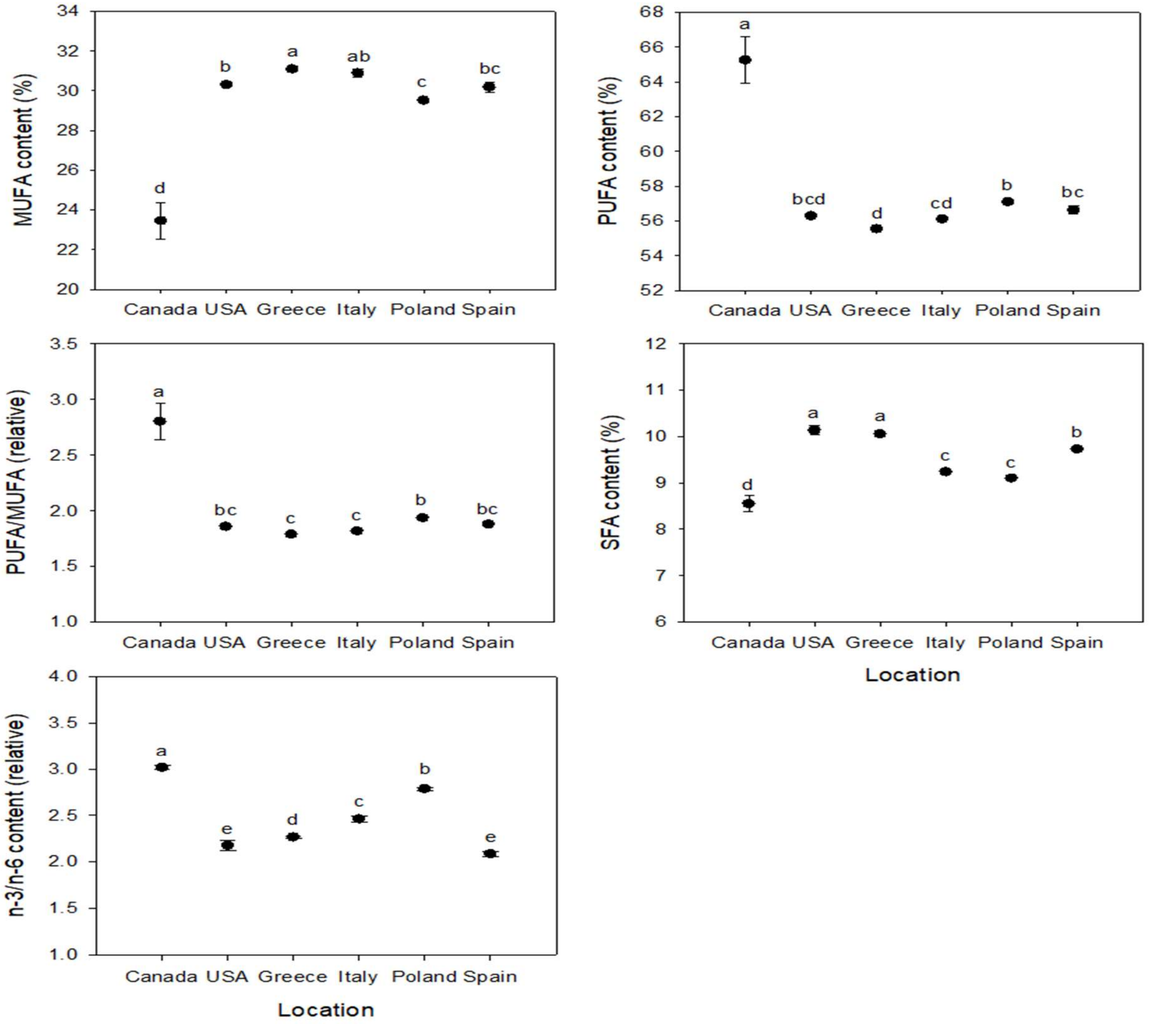
MUFA content (%)
34
32
30
28
26
24
22
20
d
b
a
ab
c
bc
Canada USA Greece Italy Poland Spain
PUFA content (%)
68
66
64
62
60
58
56
54
52
a
bcd
d
cd
b
bc
Canada USA Greece Italy Poland Spain
PUFA/MUFA (relative)
3.5
3.0
2.5
2.0
1.5
1.0
a
bc
c
c
b
bc
Canada USA Greece Italy Poland Spain
SFA content (%)
12
11
10
9
8
7
6
d
a
a
c
c
b
Canada USA Greece Italy Poland Spain
Location
n-3/n-6 content (relative)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
a
e
d
c
b
e
Canada USA Greece Italy Poland Spain
Location

Figure 3. Unsaturated FA contents (MUFA and PUFA), PUFA/MUFA ratio, n-3/n-6 ratio (ratio between omega 3 and omega 6 FAs), and Saturated (SFA) of Joelle camelina grown at each test location. Individual values are means ± standard errors. Values followed by the same letters are not statistically different for the same parameter (LSD test, *P*≤0.05).